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BROWDY AND NEIMARK, P.L.L.C.
624 NINTH STREET, NW
SUITE 300
WASHINGTON, DC 20001-5303

EXAMINER	
LEAVITT, MARIA GOMEZ	
ART UNIT	**PAPER NUMBER**
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/536,495	BRO ET AL.	
	Examiner	Art Unit	
	MARIA LEAVITT	1633	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *09 June 2008*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-9 and 11-16* is/are pending in the application.

4a) Of the above claim(s) *14-16* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-9 and 11-13* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *25 May 2005* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

Detailed Action

1. The text of those sections of Title 35, U.S. Code not included in this action can be found in a prior Office action.
2. Status of claims. Claims 1-9 and 11-16 are pending. Claims 1, 3, 6 and 7 have been amended by Applicant's amendment filed on 06-09-2008. Claims 14-16 were previously withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected invention, there being no allowable generic or linking claim. Applicant timely traversed the restriction (election) requirement in the reply filed on 12-13-2007. This application contains claim 14-16 drawn to an invention nonelected with traverse in the reply filed on 12-13-2007.
3. A complete reply to the final rejection must include cancellation of nonelected claims or other appropriate action (37 CFR 1.144) See MPEP § 821.01. The previous office action was made FINAL by the Examiner. Applicant is reminded of the right to petition under 37 CFR 1.144, if applicant disagrees with the requirements for restriction filed on 11-14-2007.

Response to Applicants' arguments in relation to restriction requirements.

The following comments are provided as a courtesy to the Applicant as the restriction requirement has already been made FINAL in the previous office action, see above.

Applicants' position at pages 9-11 of the Remarks filed on 06-09-2008 is that the disclosure of Bianchi et al., does not teach all the limitations as recited in claim 1, particularly, Applicants

argue that the "Examiner does not state where in Bianchi et al., the second feature is found". Moreover, Applicants allege that "Instead the Examiner remarks that it is unclear how the second metabolic pathway relates to the functional activity of the third enzyme and whether the first metabolite is simultaneously transformed into a second and third metabolite or they are separate metabolic conversions. Whether those matters really are unclear or not (and we say not - see below), this is clearly dodging the issue of whether in fact the second requirement numbered above is actually disclosed in Bianchi et al. Accordingly, there is no justification for the stated conclusion that the combined features are anticipated by Bianchi et al. Secondly, the Examiner newly asserts that the features in question are anticipated by Valverde et al. The Examiner explains that Valverde et al teaches expression of (we abbreviate) GAPN in E. coli which would fulfill the role of the second metabolic pathway with the increased activity of the third enzyme. However, in relation to Valverde et al, the Examiner does not explain where this teaches the first feature. Since in Valverde et al., GAPDH was deleted this is an insuperable difficulty. Thus the engineered E. coli of Valverde et al., lacks the required operative first pathway by virtue of the deletion of the NAD-dependent GAPH". As such applicants request rejoining of the claims 14-16. Such is not persuasive.

As an initial matter the current claims do not recite any first and second features. Accordingly, it is submitted that the claims fail to delineate the metes and bounds of the subject matter that Applicant regards as the invention with the requisite clarity and particularity to permit the skilled artisan to know or determine infringing subject matter as evidence by the rejection of claims 1, 6, 7 and 13 under 35 U.S.C. 112, second paragraph, in the previous office action. For the purpose of a compact prosecution, the examiner is interpreting claim 1 as having two

metabolic pathways as Applicants states at page 9 of remarks. In relation to "the engineered E. coli of Valverde et al., lacks the required operative first pathway by virtue of the deletion of the NAD-dependent GAPH", as Applicants allege, Valverde et al., discloses in preferred embodiments transformation of *E. Coli* with the GapN gene (i.e., irreversibly oxidizes G3P to 3-PGA) and consequently high production of **GAPDHN** (non-phosphorylating NADP –dependent G3P dehydrogenase or G3P:NADP oxidoreductase) (p. 153, col. 2, last paragraph; p. 155, col.1, last paragraph). Note that E. Coli comprises NAD+ dependent GAPDH. Additional *E.Coli* W3CG strains harboring three *gap-2* genes from other bacteia are disclosed at page 156, col. 1, second paragraph. Absent evidence to the contrary, the *E. coli* comprise the gapC gene encoding the cytosolic NAD-dependent **GAPDH (GAPN)** and thus comprises a first metabolic pathway wherein a first metabolite (e.g., G3P) is transformed into a second metabolite (1,3 BPGA) in a reaction in which NAD is a cofactor in addition to a second metabolic pathway "in which said first metabolite is transformed into a said further metabolite without the involvement of said second enzyme" (e.g., **GAPDHN or GAPN).**

It is noticed that the examiner has required restriction between product and process claims. Where applicant elects claims directed to the product, and a product claim is subsequently found allowable, withdrawn process claims that depend from or otherwise include all the limitations of the allowable product claim will be rejoined in accordance with the provisions of MPEP 821.04. Process claims that depend from or otherwise include all the limitations of the patentable product will be entered as a matter of right if the amendment is presented prior to final rejection or allowance, whichever is earlier. Amendments submitted after

final rejection are governed by 37 CFR 1.1 16; amendments submitted after allowance are governed by 37 CFR 1.312.

4. Therefore, claims 1-9 and 11-13 are currently being examined to which the following grounds of rejection are applicable.

Withdrawn objections/ rejections in response to Applicants' arguments or amendments:

The Requirements for Patent Applications Containing Nucleotide Sequence and/or Amino Acid Sequence Disclosures - the Sequence Rules. MPEP. 2421.03 Notification of a Failure to Comply.

In view of Applicants submission of the following requirements:

(i) reference at page 14, line 5, to the disclosed two sequences in the text of the description by use of the sequence identifier, preceded by "SEQ ID NO: 1" and by "SEQ ID NO: 2", and at page 20, lines 10, reference to the disclosed sequence by use of the sequence identifier, preceded by "SEQ ID NO: 3", and submission of a paper copy disclosing said nucleotide sequences in compliance with 37 CFR 1.821 (b)(c)(d),

(ii) submission of a copy of the "Sequence Listing" in computer readable form of the "Sequence Listing " referred to in paragraph (i) as required by 37 C.F.R. 1.821(e), and

(iii) submission of a statement at page 12 of the Remarks filed on 06-09-008 that the content of the paper and computer readable copies are the same and, where applicable, include no new matter, as required by 37 C.F.R. 1.821(e) or 1.821(f) or 1.821(g) or 1.825(b) or 1.825(d),

the notification of a Failure to Comply with the requirements of 37 CFR 1.821 through 1.825, has been withdrawn.

In addition Applicants have deleted at page 10, lines 14, the phrase "(see figure 1)", and at page 17, line 10, the phrase "Measurement of the metabolites during anaerobic fermentation is shown in Figure 3"

In view of the withdrawn objection, applicant's arguments are rendered moot.

Information Disclosure Statement

To the extent that the references have been made of record in the IDS of May 15, 2006, the discussion in the specification is considered as a voluntary comment on the relevance of the reference by Applicants.

In view of the withdrawn objection, applicant's arguments are rendered moot.

Drawings Objections

In view of Applicants' deletion at page 10, lines 14 of the phrase "(see figure 1)", and at page 17, line 10 of the phrase "Measurement of the metabolites during anaerobic fermentation is shown in Figure 3", objection to the drawings under 37 CFR 1.83(a) because they failed to show details as recited in the specification at page 10, lines 15-21 and at page 17, line 23, has been withdrawn.

In view of the withdrawn objection, applicant's arguments are rendered moot.

Specification objection

In view of Applicants deletion at page 17, line 10, of the phrase "Measurement of the metabolites during anaerobic fermentation is shown in Figure 3", objection to the disclosure because no Fig. 3 was submitted as part of the drawings filed on 05-25-2005, has been withdrawn.

In view of the withdrawn objection, applicant's arguments are rendered moot.

Claim Objection

In view of Applicants full spelling of the terms NADH, GAPN and GAPDH, objection to claims 1, 6, and 7, has been withdrawn. Note that Applicants suggest spelling out the abbreviation first followed by the corresponding abbreviation in parenthesis. The latter is the customary description of abbreviations at the first encounter in the claims.

In view of the withdrawn objection, applicant's arguments are rendered moot.

Rejections maintained in response to Applicants' arguments or amendments

Claim Rejections - 35 USC § 112 (second paragraph)

Claims 1, 6, 7 and 13 remain rejected under 35 U.S.C. 112, second paragraph, as being indefinite in that it fails to point out what is included or excluded by the claim language.

Response to Applicants' Arguments as they apply to rejection of claims 1, 6, 7 and 13 ***under 35 USC § 112 (second paragraph.)***

In relation to the rejection of claim 1 as being vague and indefinite in its recitation of the phrase "an operative second metabolic pathway characterised by an enzyme activity of a native level in respect of a third enzyme catalysing a non-reversible reaction" , Applicants allege

at page 14 of Remarks, that the interpretation of the term "native level" by the Examiner was not intended. Moreover, Applicants have deleted the word "in respect" to make clear the meaning of the term "native level". As such, Applicants state, " What is intended is of course that there should be above a native level of activity of the third enzyme. In the illustrated case, the native activity of the third enzyme in the yeast is zero, because the native form of the yeast does not express GAPN. The wording is also apt to cover a situation in which the third enzyme activity is natively present, but has been increased such as by over expression". Such is not persuasive.

The examiner notes that Applicants are arguing limitations not present in claim 1. Hence, it is not possible to know the metes and bounds of " an enzyme activity in excess of a native level of a third enzyme catalysing a non-reversible reaction " because it is unclear whether the native activity of a third enzyme refers to a wild type enzyme genetically modified or not, what is the comparative reference so as to define how much is "in excess", is the excess relative to a value of zero of a third enzyme, as applicants contend, or is there an enhanced activity because "a third" enzyme has been overexpressed? . The phrase "an enzyme activity in excess of a native level of a third enzyme catalysing a non-reversible reaction" is indefinite because the specification does not clearly define the phrase. Thus the metes and bounds are of the claim are unclear.

In relation to the transformation of a first metabolite into a second metabolite and the transformation of said second metabolite into at least one further metabolite "in which said first metabolite is transformed into a said further metabolite without the involvement of said second enzyme" as recited in claim 1 ", Applicants argue to be confused by the rejection of the claim based on whether the metabolic transformation take place separately or simultaneously, as the

Examiner pointed out. Applicants contend that "Clearly these transformations of the first metabolite into the second and third metabolites are 'separate' because they start from the first metabolite and go to different products. Equally, they are expected to be "simultaneous" in that both processes will be ongoing at the same time. The dichotomy proposed by the Examiner does not seem to us to exist". Such is not persuasive.

As set forth in the previous office action, it is unclear as to whether the claims are intended to be limited to a metabolically engineered microorganism wherein the transformation of the first metabolite to the second metabolite and transformation of said second metabolite into at least one further metabolite takes place simultaneously with the transformation of what appears to be a second metabolic pathway, i.e., "said first metabolite is transformed into a said further metabolite without the involvement of said second enzyme", or the conversion of said second metabolic pathway takes place sequentially after the first conversion of the first metabolite to the second and to the "at least one further metabolite". Note that the second enzyme is not active in what appears to be a second metabolic pathway.

In so far as the use of identification numbers in parenthesis for GAPN in claims 6 and 13, and GAPDH in claim 7, Applicants allege that "we believe it to be standard practice to use parentheses to give an alternative name for a material, for instance where initials are followed by the full name of a compound or vice versa. The EC number provides the standard classification and thus helps to clarify which enzyme is intended for the enzyme in question. In many cases, the EC number refers to a single enzyme". Such is not persuasive.

The specification as filed does not disclose the meaning of "GAPN (EC 1.2.1.9" and "GAPDH (EC 1.2.1.12)" . However, at page 15, lines 10 -15, the specification teaches the

overall conversion of glucose to the different metabolites for the GAPN strain. Hence it is unclear whether the EC number generically identifies all the GAPN strains or is strain specific, e.g., *Streptococcus, S. cerevisiae* and others. Accordingly, it is submitted that the claims fail to delineate the metes and bounds of the subject matter that Applicant regards as the invention with the requisite clarity and particularity to permit the skilled artisan to know or determine infringing subject matter.

Claim Rejections - 35 USC § 103

Claims 1-9 and 11-13 remain rejected under 435 U.S.C. 103(a) as being unpatentable over of Nissen et al., (Metabolic Engineering, 2000, 2: pages 69-77) in view of Valverde et al., (FEBS, 1999, 21898, pages 153-158).

Response to Applicants' Arguments as they apply to rejection of claims 1-9 and 11-13 ***under 35 USC § 103***

At page 17 of Remarks, Applicants' position is that the Examiner starts from a false premise. Specifically, Applicants state, "Nissen et al' s teaching is significantly misstated. It does not teach reduction in the production of NADH. Rather, it teaches provision of an NADH consuming reaction path that does not lead to glycerol. This undermines the Examiner's argument, which starts from a false premise"[emphasis added]. Such is not persuasive.

As an initial matter, the Examiner notes that during alcoholic fermentation by yeast, the mayor role of glycerol formation is to maintain the redox balance. While ethanol production which ensures reoxidation of the NADH (e.g., NADH + H+ $\rightarrow$ NAD+) formed during oxidation of Glyceraldehyde 3-phosphate, is a redox equilibrated process, excess NADH is produced during biomass formation and secondary fermentation products. Glycerol is mainly produced to

counter balance this surplus of NADH and may be considered to form a redox valve (Michnick et al., 1997, Yeast, 13:783-793; p. 783, col. 2, of record; Nissen, p. 69, Abstract). Indeed, Nissen et al., generates a modified yeast to reduce the surplus of NADH, formed by the synthesis of biomass and secondary fermentation, by replacing the normal NADPH-consuming synthesis of glutamate dehydrogenase (e.g., glutamate dehydrogenase was deleted) with a new pathway in which ATP and NADH were consumed. Therefore, the total surplus of NADH in a fermentation reaction was reduced. While the pathway modified by Nissen et al., is involved in biomass synthesis (e.g., ammonium ion incorporation into glutamate) and not necessarily in the synthesis of glycerol, the total reduction of NADH surplus will contribute to the redox balance of NAD+/NADH in the fermentation of yeast as well as the reoxidation of NADH to NAD+. Note that fermentation preferentially requires anaerobic conditions, under anaerobic conditions NADH is reoxidized to NAD+ by formation of glycerol from glucose. Hence, Examiner's argument does not starts from a false premise.

At page 17 of remarks, Applicants contend that "the metabolic engineering in Valverde et al is that the resulting *E. coli* is unable to grow anaerobically, the very problem that Valverde et al were trying to avoid. A skilled reader would therefore not perceive Valverde et al as offering a teaching likely to be useful in yeast as an alternative strategy for obtaining the objects of Nissen et al (reduced glycerol and increased ethanol)". Moreover, Applicants argue that "The two teachings lie in very different fields, one being concerned with the metabolism of a yeast and the other with the metabolism of a bacteria. A skilled person would be unlikely to look for an alternative solution to the problem addressed in Nissen et al relating to yeast in a teaching confined to the metabolism of E. coli metabolic engineering in Valverde et al is that the resulting

E. coli is unable to grow anaerobically, the very problem that Valverde et al were trying to avoid. A skilled reader would therefore not perceive Valverde et al as offering a teaching likely to be useful in yeast as an alternative strategy for obtaining the objects of Nissen et al (reduced glycerol and increased ethanol).

At the outset, the examiner notes that the instant claims do not recite any limitation related to an anaerobic or aerobic fermentative process. In contrast to Applicants arguments, Nissen teaches optimization of ethanol production in yeast which preferentially requires anaerobic conditions. As stated in the paragraph above, NADH can be reoxidized under aerobic conditions, but this demands critical control of oxygen level to maintain fermentative metabolism and ethanol production. As fermentation may take place under aerobic and anaerobic conditions, there is not evidence why one of ordinary skill in the art, on teachings provided by the combined cited references, would not have been motivated to employ the non-phosphorylating NADP-dependent G3P dehydrogenase (GAPDHN or GAPN) as taught by Valverde in the fermentation process of Nissen with the goal of reducing the total NADH surplus so as to increase ethanol production and to reduce glycerol yield. Furthermore, the Valverde discloses *E. coli gapC* mutant, e.g., W3CG, transformed with a functional plant GAPDHN. There is not reason why a functional pea plant GAPDHN couldn't be expressed in transformed *Yeast* with a recombinant DNA molecule as easily as in *E. Coli* to obtain similar benefits.

At pages 17 and 18 of Remarks, Applicants argue that "a skilled reader would not have found it obvious to combine the teachings as proposed and would not have had a reasonable expectation that the combination would should have an intact GAPDH which produces NADH, whereas in Valverde et al, the E. coli had its NAD-dependent GAPDH deleted. We submit that if

it were obvious to combine the teachings of Nissen et al and Valverde et al at all (which we deny), the result would be a yeast in which, per Valverde et al, GAPDH would be deleted and GAPN would be introduced. That however would not meet the requirements of claim 1. The Examiner cannot properly pick and choose which features of Nissen and Valverde to combine on the basis of applicant's own teachings; that is hindsight reconstruction. Yet the Examiner fails to articulate any teaching in Nissen which would direct the ordinary worker to pick only the features of Valverde which the Examiner has emphasized" [emphasis added]. Such is not persuasive.

While Valverde discloses transformation of *E. coli gapC* mutant, e.g., W3CG, transformed with a functional plant GAPDHN, Valverde additionally teaches transformation of *E. Coli* with the GapN gene (i.e., irreversibly oxidizes G3P to 3-PGA) and consequently high production of plant **GAPDHN** or GAPN (non-phosphorylating NADP –dependent G3P dehydrogenase or G3P:NADP oxidoreductase) in the presence of the IPTG inducer (p. 153, col. 2, last paragraph; p. 155, col.1, last paragraph). Note that E. coli comprises endogenous GAPH (GAPDH). Additionally, Valverde teaches *E.Coli* W3CG strains harboring three *gap-2* genes from other bacteria are disclosed at page 156, col. 1, second paragraph. Absent evidence to the contrary, the *E. coli* transformant comprises both the phosphorylating and non-phosphorylating branches of glycolysis (p. 157, Fig. 4). Clearly, the teachings of Valverde et al., complement the teachings of Nissen in relation to metabolically engineered fermentative microorganisms and glucose catabolism. Expression of the **GAPDHN** could potentially represent an alternative metabolic route in Yeast to reduce excess NADH produced during biomass formation and secondary fermentation products and thus increase the ethanol yield in yeast. Additionally, the

examiner notices that KSR forecloses the argument that a specific teaching, suggestion, or motivation is required to support a finding of obviousness. See the recent Board decision *Ex parte Smith*, --USPQ2d--, slip op. at 20, (Bd. Pat. App. & Interf. June 25, 2007) (citing KSR, 82 USPQ2d at 1396).

At page 18 of Remarks, Applicants further argue that Nissen et al., discloses a way to "drain off" surplus production of NADH that will otherwise lead to glycerol production, no such draining off mechanism is taught by Valverde. Accordingly, Applicants allege, the proposed combination is not a substitution of like and like. "Instead, Valverde et al teaches that the ability to metabolize sugar lost via a deletion of GAPDH can be partially rescued (but only under aerobic conditions) by introducing GAPN. According to the Examiner, it is apparent that the catabolic yield of GAPN includes NADPH. This is clearly quite different from draining off NADH". Such is not persuasive.

The final outcome in the metabolically engineered Yeast taught by Nissen is, in part, increased consumption of ATP and NADH, thus reducing the total surplus of NADH during fermentation. The final outcome of the metabolically engineered *E. Coli* disclosed by Valverde is, in part, recovery of the inoperative phosphorylating branch of glucose catabolism due to a mutation of NAD+ -dependent glyceraldehyde 3-phosphatase. Thus transformant *E. coli* W3CG with pea GAPDHN (GAPN) plant, generated a functional pathway for the bacteria to grow aerobically on sugars but failing to perform anaerobic sugar fermentation or aerobic growth. GAPDHN catalyses the irreversible oxidation of glyceraldehyde-3-phosphate and $NADP^+$ into 3-phosphoglycerate and NADPH. The reaction catalysed by GAPDHN (GAPN) yields one NADPH instead of one NADH and one ATP when comparing with the total reaction catalysed

by **NAD^+-dependent glyceraldehyde-3-phosphate dehydrogenase (GAPDH)** and phosphoglycerate kinase (PGK). Clearly, by transforming a Yeast with a plasmid encoding GAPDHN (GAPN), wherein the *gapC* gene encoding GAPDH is mutated, it should be reasonably expected to convert G3P into 3-PGA by yielding one NADPH and not one NADH and one ATP molecules.

Additionally, at pages 18 and 19 of Remarks, Applicants raise the following issues: (i) It would have been unknown whether GAPN could be expressed successfully in yeast, (ii) it would have been unknown whether production of NADPH via GAPN would have had any substantial effect on the level of NADH in yeast, it would have been unknown to what extent GAPN if expressed successfully in yeast would become engaged in glycolysis competing with the native yeast enzymes, (iii), the consequence of metabolizing G3P to 3-PGA via GAPN rather than via GAPDH is that one does not get production of ATP. However, the effect on the production of glycerol and ethanol in yeast of this loss of ATP production would have been quite unknown. "As seen in the diagram on page 157 of Valverde et al, ATP is required for consumption in the earlier stages of metabolism of glucose. Valverde et al had reported that their engineered E. coli had a decreased growth rate compared to wild type". Such is not persuasive.

As set forth in the paragraph above, there is no reason why a functional p**ea plant GAPDHN** (GAPN) couldn't be expressed in transformed *Yeast* with a recombinant DNA, (ii) once again, microorganisms need to oxide the excess NADH by achieving a redox balance. Excess NADH is reoxidized either by respiration or formation of unwanted side products such as glycerol. Nissen clearly discloses that reduction of NADH and ATP surplus reduces glycerol yield and increases ethanol production, Valverde complements the teachings of Nissen by

disclosing that *E. coli* transformants with a modified glycolytic pathway wherein direct production of 3-PGA from G3P is achieved bypassing the ATP generated reaction and using NADP+ as cofactor rather than NAD+. Valverde demonstrated that avoiding one route of production of NADH by expressing a plant GAPN, functionality of the W3CG mutant, which lacks GAPDH is recovered, albeit with different growth and substrate conversion. For example, Valverde teaches that complementation of the mutant endogenous GAPDH from three clones harboring recombinant GAPDHs, the unicellular cyanobacterium *Synechocystis,* the filamentous cyanobacterium *Anabaena and E. coli* , are able to fermentate sugars and grow on gluconeogenic substrates (acetate plus succinate) as the sole carbon sources (p. 156, col. 1, second paragraph). Thus, Valverde extensively evidence how a heterologous GAPN is successfully expressed in *E. coli* and functionally recovered the glycolic pathway. Furthermore, the plant remains functional and its functionality is completed by expression of other recombinant GAPDHs in the same mutant *E. coli.*

Conclusion

Claims 1-9 and 11-13 are not allowed.

THIS ACTION IS MADE FINAL. Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period

will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Maria Leavitt whose telephone number is 571-272-1085. The examiner can normally be reached on M-F.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Joseph Woitach, Ph.D can be reached on (571) 272-0739. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

To aid in correlating any papers for this application, all further correspondence regarding his application should be directed to Group Art Unit 1636; Central Fax No. (571) 273-8300. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to (571) 272-0547.

Patent applicants with problems or questions regarding electronic images that can be viewed in the Patent Application Information Retrieval system (PAIR) can now contact the USPTO's Patent Electronic Business Center (Patent EBC) for assistance. Representatives are available to answer your questions daily from 6 am to midnight (EST). The toll free number is (866) 217-9197. When calling please have your application serial or patent number, the type of document you are having an image problem with, the number of pages and the specific nature of the problem. The Patent Electronic Business Center will notify applicants of the resolution of the problem within 5-7 business days. Applicants can also check PAIR to confirm that the problem has been corrected. The USPTO's Patent Electronic Business Center is a complete service center supporting all patent business on the Internet. The USPTO's PAIR system provides Internet-based access to patent application status and history information. It also enables applicants to view the scanned images of their own application file folder(s) as well as general patent information available to the public.

Maria Leavitt, PhD
Patent Examiner P/1633
Remsen 2B55
Phone: 571-272-1085

/Anne Marie S. Wehbe/

Primary Examiner, Art Unit 1633